Null Results in Brief

Serum Insulin-like Growth Factor I (IGF-I) Concentration in Men Is Not Associated with the Cytosine-Adenosine Repeat Polymorphism of the *IGF-I* Gene[1]

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Introduction

IGF-I[3] is a polypeptide involved in the control of mitogenesis, cell-cycle regulation, and cell survival. Prospective epidemiological studies suggest that elevated levels of IGF-I, either as absolute concentrations or relative to levels of insulin-like binding protein-3 (IGFBP-3), are a risk factor for the development of several common types of cancer (1). IGF-I levels vary substantially between individuals, and it has been estimated that up to 60% of the between-person variability has a genetic basis (2), although the specific loci involved are unknown. A microsatellite polymorphism, comprising a variable length CA repeat sequence, has been identified in the promoter region of the *IGF-I* gene, 1 kb upstream from the IGF-I transcription initiation site, a region that contains specific regulatory agents (3). The functional significance of this polymorphism is not yet known, although it may alter promotional activity and, thus, influence the transcription rate of IGF-I. The number of repeat units in the CA polymorphism range from 11 to 24 repeats; the most common allele has 19 repeats (*CA19*). In a previous study of 116 white men and women, the homozygous *CA19/CA19* genotype was associated with a 16% lower serum IGF-I concentration than were other genotypes (3). Another study found 78 black American women to have a lower prevalence of the *CA19* allele than did 329 Caucasian women, as well as higher mean circulating IGF-I levels (4), which parallels their increased breast cancer risk. However, among these premenopausal women, an association between the *CA19* allele and IGF-I levels was reported only among users of oral contraceptives (4). The aim of this study was to test the hypothesis that the 19-repeat allele of the *IGF-I* gene (*CA19*) is associated with a lower circulating IGF-I concentration in Caucasian men.

Materials and Methods

This study is based on a sample of 696 men recruited from the Oxford, United Kingdom component of EPIC. All of the men were of Caucasian origin with a mean age of 47 years (range, 20–78 years); a detailed description of subject recruitment and inclusion criteria is published elsewhere (5). DNA was purified from 0.5 ml of buffy coat samples of peripheral blood using Nucleon BACC2 kits according to the manufacturer's instructions (Nucleon ST, Glasgow, Scotland). Genotyping was completed using a PCR-RFLP method as described by Jernström *et al.* (4). The number of repeat elements was determined and genotypes were defined according to the presence or absence of a *CA19* repeat allele. Measurements of serum IGF-I concentration were performed at the Clinical Biochemistry Laboratory at the John Radcliffe Hospital, Oxford, as described previously (5). IGF-I concentrations were square-root transformed to approximate a normal distribution; all mean values and corresponding 95% confidence intervals are presented as back-transformed values. The mean IGF-I concentration in each genotype was calculated using analysis of covariance after adjustment for age, body mass index [weight (kg)/height (m^2)], dietary group, smoking status, and days between venipuncture and blood processing. All of the statistical analyses were performed using Stata version 5.0.

Results

Genotyping was successful for 660 (95%) of 696 study participants. The CA repeat sequences ranged from 11 to 24, although *19CA* repeats were the most common, with an allele frequency of 64%. Two hundred and seventy (40.9%) subjects

Table 1 Mean IGF-I concentration (nmol/liter) according to the *IGF-I 19CA* repeat allele

IGF-I 19CA repeat allele	No. of participants	Mean (95% CI[a]) IGF-I[b]
Two *19CA* alleles	270 (41%)	19.7 (19.1–20.4)
One *19CA* allele	308 (47%)	19.6 (19.0–20.2)
No *19CA* alleles	82 (12%)	19.5 (18.3–20.6)
Test for heterogeneity		$P = 0.906$
Test for linear trend		$P = 0.657$
19CA present	578 (88%)	19.6 (19.2–20.1)
19CA absent	82 (12%)	19.5 (18.3–20.6)
Test for heterogeneity		$P = 0.761$

[a] CI, confidence interval.
[b] All of the values are adjusted for age (20–24, . . . , 65+), body mass index (<20, 20–21.9, 22–23.9, 24–25.9, 26–27.9, 28–29.9, 30+ kg/m^2), dietary group (meat-eater, vegetarian, vegan), smoking status (never, past, 1–9 cigarettes/day or pipe or cigar smoker, 10+ cigarettes/day), and days between venipuncture and blood processing (1, 2, 3, 4+ days).

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[3] The abbreviations used are: IGF-I, Insulin-like growth factor-I; CA, cytosine-adenosine; EPIC, European Prospective Investigation into Cancer and Nutrition.

had two *CA19* repeat alleles, 308 (46.7%) had one *CA19* repeat allele, and 82 (12.4%) had no *CA19* repeat alleles. The genotype frequencies were in Hardy-Weinberg equilibrium ($P = 0.95$) and are consistent with previous data (4). We found no evidence to suggest that the presence of the *CA19* repeat allele was associated with a lower (or higher) serum IGF-I concentration, either before or after adjustment for possible confounders (Table 1). Similarly, there was no trend between increasing length of the repeat allele and IGF-I concentration (data not shown).

Discussion

This cross-sectional study is the largest to date to investigate a possible association between the *IGF-I CA19* repeat sequence and circulating IGF-I concentrations. The study had 80% power to detect a 7% difference in IGF-I between genotypes. Limitations included the assumption that a single measurement of IGF-I is an accurate reflection of long-term IGF-I status in men and the relatively low numbers of subjects without a *CA19* allele. Although a previous study found the *CA19* allele to be associated with a significantly lower IGF-I concentration among Caucasian men and women (3), our data suggest that the *IGF-I CA19* polymorphism is not associated with circulating IGF-I concentrations in adult men. Given the large heritable component in the variation of IGF-I levels, it is possible that other variants in the *IGF-I* gene are associated with IGF-I concentrations and, possibly, subsequent cancer risk.

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